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APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
10/053,526	01/18/2002	Marie Dutreix	3754/OK213	1617

7590 07/24/2003

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EXAMINER
FREDMAN, JEFFREY NORMAN

ART UNIT	PAPER NUMBER
1634	

DATE MAILED: 07/24/2003

Please find below and/or attached an Office communication concerning this application or proceeding.

Office Action Summary	Application No.	Applicant(s)	
	10/053,526	DUTREIX ET AL.	
	Examin r	Art Unit	
	Jeffrey Fredman	1634	

-- The MAILING DATE of this communication app ars on the cov r she t with th correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If the period for reply specified above is less than thirty (30) days, a reply within the statutory minimum of thirty (30) days will be considered timely.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133).
- Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *02 June 2003* .

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1-23* is/are pending in the application.

4a) Of the above claim(s) *22* is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☐ Claim(s) _____ is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

11)☐ The proposed drawing correction filed on _____ is: a)☐ approved b)☐ disapproved by the Examiner.

If approved, corrected drawings are required in reply to this Office action.

12)☐ The oath or declaration is objected to by the Examiner.

Priority under 35 U.S.C. §§ 119 and 120

13)☒ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☒ None of:

1.☒ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____ .

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

14)☐ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. § 119(e) (to a provisional application).

a) ☐ The translation of the foreign language provisional application has been received.

15)☐ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. §§ 120 and/or 121.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☒ Information Disclosure Statement(s) (PTO-1449) Paper No(s) _____ .
4) ☐ Interview Summary (PTO-413) Paper No(s). _____ .
5) ☐ Notice of Informal Patent Application (PTO-152)
6) ☐ Other:

DETAILED ACTION

Election/Restrictions

1. Applicant's election with traverse of Group I in the paper filed June 2, 2003, is acknowledged. The traversal is on the ground(s) that these are biotechnological process and product and should therefore be examined together. This is not found persuasive because 35 U.S.C. 103(b) is designed for the situation where the product is found to be novel and unobvious, not the method. Had Applicant elected Group II and Group II been found novel, then rejoinder would have been appropriate under 103(b). In the current case, where the method claim was elected, there is no expectation that simply because a method is allowable (which currently the method is not), the product would also be allowable. Therefore, the restriction is maintained.

The requirement is still deemed proper and is therefore made FINAL.

Claim Rejections - 35 USC § 112

2. Claims 2, 4, 8, 10, 12 and 14 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

In claim 2, it is vague and indefinite what is meant by "donor nucleic acid is prepared by chemical synthesis **of** by an amplification method (my emphasis)." The language of this claim is unclear. For prior art purposes, it will be assumed that "of" is supposed to be "or".

In claim 4, it is vague and indefinite how a "double stranded nucleic acid" can interact by Watson-Crick base pairing to the adaptor nucleic acid when the double stranded nucleic acid has no single stranded regions.

In claims 8 and 10, the phrase "preferably" renders the claim indefinite because it is unclear whether the limitations following the phrase are part of the claimed invention. See MPEP § 2173.05(d).

In claim 12, it is vague and indefinite what is meant by the word "optionnaly".

In claim 14, the word "hexaethyleglycol" is indefinite. If "hexaethyleneglycol" is meant, correction is required.

Claim Rejections - 35 USC § 102

3. The following is a quotation of the appropriate paragraphs of 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

> A person shall be entitled to a patent unless –
>
> (b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

4. Claims 1, 2, 4, 8-13, 15-21 and 23 are rejected under 35 U.S.C. 102(b) as being anticipated by Chan et al (J. Biol. Chem. (April 1999) 274:11541-11548).

As an initial matter, the claims must be interpreted to determine how the prior art applies. Here, the required elements are a TFO oligonucleotide sequence which is linked to a sequence that hybridizes by Watson-Crick hybridization to another sequence, following which the TFO sequence binds to a cellular nucleic acid and a recombination event using the attached sequences occurs.

Chan teaches a method for effecting a homologous recombination between a native nucleic acid segment and a donor segment introduced into a cell (see abstract) comprising:

(a) introducing into a cell a nucleic acid targeting system (see page 11542, columns 1 and 2) comprising:

i) a third strand oligonucleotide which comprises a base sequence that forms a triple helix at a binding region on a native nucleic acid segment (see page 11543, figure 1, triplex forming domain and page 11542, column 1),

ii) an adaptor segment comprising an oligonucleotide able to bind at least a portion of the donor nucleic acid by Watson Crick base pairing which is linked to the third strand oligonucleotide (see page 11543, figure 1, where there is a oligonucleotide linked by a linker to the triplex forming domain. This linked oligonucleotide is deemed to be the adaptor segment for purposes of the rejection),

iii) a donor nucleic acid comprising a nucleic acid sequence homologous to the native nucleic acid segment so that the donor sequence is capable of undergoing homologous recombination with the native sequence at the target region (see page 11543, figure 1, where there is a single strand hybridized to the "donor" domain (here adaptor sequence) that is linked to the TFO oligonucleotide, and this single strand is capable of undergoing homologous recombination to repair the native sequence).

(b) allowing the third strand oligonucleotide to bind to the native nuclei acid segment to form a triple helix nucleic acid, thereby inducing homologous recombination

at the native nucleic acid target region (see page 11543, figure 1, figure 2 and page 11542, subheading "In vitro triplex formation" and "intracellular targeting protocol"),

(c) allowing homologous recombination to occur between the native and donor nucleic acid segments (see page 11543, figure 1, figure 2 and page 11542, subheading "In vitro triplex formation" and "intracellular targeting protocol").

With regard to claim 2, Chan teaches preparation of the oligonucleotide by chemical synthesis (see page 11542, subheading "oligonucleotides").

With regard to claim 4, Chan teaches a double and single stranded donor nucleic acid (see page 11543, figure 1).

With regard to claim 8, Chan teaches using a 30 nucleotide TFO region (see page 11543, AG30 domain used as TFO region).

With regard to claim 9, Chan teaches using an approximately 40mer nucleic acid as donor (see page 11543, figure 2).

With regard to claim 10, Chan teaches the use of an adaptor that is about 40 mer nucleic acid (see page 11543, figure 2).

With regard to claims 11-13, Chan teaches the use of a polyethyleneglycol linker (see 11544, figure 3 and page 11543, figure 2).

With regard to claims 15-21, Chan teaches correction of a mutation including base changes in an extrachromosomal, plasmid type, sequence (see page 11543, figure 2). Chan expressly recognizes the applicability to chromosomal correction (see page 11548, column 2).

Claim Rejections - 35 USC § 103

5. The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

6. This application currently names joint inventors. In considering patentability of the claims under 35 U.S.C. 103(a), the examiner presumes that the subject matter of the various claims was commonly owned at the time any inventions covered therein were made absent any evidence to the contrary. Applicant is advised of the obligation under 37 CFR 1.56 to point out the inventor and invention dates of each claim that was not commonly owned at the time a later invention was made in order for the examiner to consider the applicability of 35 U.S.C. 103(c) and potential 35 U.S.C. 102(e), (f) or (g) prior art under 35 U.S.C. 103(a).

7. Claims 1-5, 8-13, 15-21 and 23 are rejected under 35 U.S.C. 103(a) as being unpatentable over Chan et al (J. Biol. Chem. (April 1999) 274:11541-11548) in view of Erdeniz et al (Genome Research (1997) 7:1774-1183).

Chan teaches a method for effecting a homologous recombination between a native nucleic acid segment and a donor segment introduced into a cell (see abstract) comprising:

(a) introducing into a cell a nucleic acid targeting system (see page 11542, columns 1 and 2) comprising:

i) a third strand oligonucleotide which comprises a base sequence that forms a triple helix at a binding region on a native nucleic acid segment (see page 11543, figure 1, triplex forming domain and page 11542, column 1),

ii) an adaptor segment comprising an oligonucleotide able to bind at least a portion of the donor nucleic acid by Watson Crick base pairing which is linked to the third strand oligonucleotide (see page 11543, figure 1, where there is a oligonucleotide linked by a linker to the triplex forming domain. This linked oligonucleotide is deemed to be the adaptor segment for purposes of the rejection),

iii) a donor nucleic acid comprising a nucleic acid sequence homologous to the native nucleic acid segment so that the donor sequence is capable of undergoing homologous recombination with the native sequence at the target region (see page 11543, figure 1, where there is a single strand hybridized to the "donor" domain (here adaptor sequence) that is linked to the TFO oligonucleotide, and this single strand is capable of undergoing homologous recombination to repair the native sequence).

(b) allowing the third strand oligonucleotide to bind to the native nuclei acid segment to form a triple helix nucleic acid, thereby inducing homologous recombination at the native nucleic acid target region (see page 11543, figure 1, figure 2 and page 11542, subheading "In vitro triplex formation" and "intracellular targeting protocol"),

(c) allowing homologous recombination to occur between the native and donor nucleic acid segments (see page 11543, figure 1, figure 2 and page 11542, subheading "In vitro triplex formation" and "intracellular targeting protocol").

With regard to claim 2, Chan teaches preparation of the oligonucleotide by chemical synthesis (see page 11542, subheading "oligonucleotides").

With regard to claim 4, Chan teaches a double and single stranded donor nucleic acid (see page 11543, figure 1).

With regard to claim 8, Chan teaches using a 30 nucleotide TFO region (see page 11543, AG30 domain used as TFO region).

With regard to claim 9, Chan teaches using an approximately 40mer nucleic acid as donor (see page 11543, figure 2).

With regard to claim 10, Chan teaches the use of an adaptor that is about 40 mer nucleic acid (see page 11543, figure 2).

With regard to claims 11-13, Chan teaches the use of a polyethyleneglycol linker (see 11544, figure 3 and page 11543, figure 2).

With regard to claims 15-21, Chan teaches correction of a mutation including base changes in an extrachromosomal, plasmid type, sequence (see page 11543, figure 2). Chan expressly recognizes the applicability to chromosomal correction (see page 11548, column 2).

Chan does not teach preparation of nucleic acid for recombination by PCR amplification.

Erdeniz teaches preparation of nucleic acid for recombination (see abstract) comprising the steps:

(a) providing a pair of primers complementary to a target native sequence (see page 1181, table 2),

(b) amplifying said first nucleic acid sequence (see page 1181, subheading "PCR"),

(c) isolating the amplification thus obtained (see page 1182, column 2),

(d) initating a recombination event (see page 1177, figure 3, for example).

It would have been prima facie obvious to one of ordinary skill in the art at the time the invention was made to synthesize the nucleic acid constructs of Chan by use of the PCR reaction as taught by Erdeniz since Erdeniz states "The advantages of these methods are that any mutation can be created easily using PCR primers without any need for cloning (see page 1174, column 2 to page 1175, column 1)." Erdeniz also recognizes that these approaches have some drawbacks, most notably low integration frequency (see page 1175, column 2)." Thus, an ordinary practitioner, who wished to take advantage of the ease of mutation generation by PCR but who wished to avoid the cost of low integration frequency would have been motivated to link the PCR generated fragment to a TFO oligonucleotide as taught by Chan since Chan expressly notes that use of the TFO domain increases recombination by up to 50 fold (see abstract).

8. Claims 1, 2, 4, 8-21 and 23 are rejected under 35 U.S.C. 103(a) as being unpatentable over Chan et al (J. Biol. Chem. (April 1999) 274:11541-11548) in view of Sato et al (U.S. Patent 5,770,408).

Chan teaches the limitations of claims 1, 2, 4, 8-13, 15-21 and 23 as discussed above. Chan does not teach the use of a hexaethyleneglycol linker.

Sato teaches the use of a hexaethyleneglycol linker (see column 6, lines 19-21).

It would have been prima facie obvious to one of ordinary skill in the art at the time the invention was made to synthesize the nucleic acid constructs of Chan by use of the hexaethyleneglycol linker of Sato since Sato states that the hexaethyleneglycol linker is a desirable linker. Further, an ordinary practitioner would select this linker because it is nuclease resistant as well as being an equivalent linker to the linker used by Chan. MPEP 2144.06 notes " Substituting equivalents known for the same purpose. In order to rely on equivalence as a rationale supporting an obviousness rejection, the equivalency must be recognized in the prior art, and cannot be based on applicant's disclosure or the mere fact that the components at issue are functional or mechanical equivalents. An express suggestion to substitute one equivalent component or process for another is not necessary to render such substitution obvious. In re Fout , 675 F.2d 297, 213 USPQ 532 (CCPA 1982)." Here, the hexaethyleneglycol linker is a known equivalent to the linker of Chan.

Claim Rejections - 35 USC § 112

9. The following is a quotation of the first paragraph of 35 U.S.C. 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

10. Claims 1-21 and 23 are rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for in vitro site directed mutagenesis of a target DNA molecule or site directed mutagenesis of a target DNA molecule ex vivo in cultured or isolated cells, but does not reasonably provide enablement for in vivo methods of site

directed mutagenesis of a target DNA molecule. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to use the invention commensurate in scope with these claims.

Factors to be considered in determining whether a disclosure meets the enablement requirement of 35 USC 112, first paragraph, have been described by the court in *In re Wands*, 8 USPQ2d 1400 (CA FC 1988). *Wands* states at page 1404,

> "Factors to be considered in determining whether a disclosure would require undue experimentation have been summarized by the board in Ex parte Forman. They include (1) the quantity of experimentation necessary, (2) the amount of direction or guidance presented, (3) the presence or absence of working examples, (4) the nature of the invention, (5) the state of the prior art, (6) the relative skill of those in the art, (7) the predictability or unpredictability of the art, and (8) the breadth of the claims."

The nature of the invention and breadth of claims

Claims 1-21 and 23 are broadly drawn to methods of site directed mutagenesis comprising a nucleic acid linked to a single stranded nucleic acid that forms a triple helix with the target region which encompasses in vivo, ex vivo and in vitro methods. In fact the specification recites that the present invention provides in vivo and in vitro site directed mutagenesis of a target DNA molecule. However, as will be further discussed, there is no support in the specification and prior art for the in vivo methods, only for ex vivo or in vitro methods. The invention is an class of invention which the CAFC has characterized as "the unpredictable arts such as chemistry and biology." Mycogen Plant Sci., Inc. v. Monsanto Co., 243 F.3d 1316, 1330 (Fed. Cir. 2001).

The unpredictability of the art and the state of the prior art

The specification recites site specific, targeted mutagenesis of the gene in an ex vivo method (see specification, example 6). However, there is no evidence that said site-directed mutagenesis method would be operable in vivo. In example 6 of the specification, culture CHO cells were site directed mutated by the oligonucleotide mutagen complex added to the growth medium. However, there is no correlation between the entry of the oligonucleotide-mutagen complex in isolated cells in an ex vivo method and in vivo applications where entry into an animal is required.

There is a great deal of unpredictability in the modulation of nucleic acid interactions in vivo. Similar problems are also faced by ribozyme therapy. Uhlmann et al. (Chem. Reviews 90 : 544-584 (1990)) teach that the secondary and tertiary structure of the target nucleic acids have a critical influence on the efficiency of the target site and that it is impossible to predict the higher order structure of the mRNA and the effect it will have on the efficacy of any potential inhibitory oligo (p. 576). Mirabelli et al (Anticancer Drug Design 6 :647-661 (1991)) teaches that we do not currently understand the precise role of nucleases, other intracellular enzymes and proteins on the stability of the ribozymes, the process by which oligonucleotides penetrate cellular membranes and distribute in cells, the non-sequence-specific-interactions that oligonucleotides can engage in both in and out of cells, and the metabolic pathways (both nuclease and non-nuclease) and metabolites that are likely to play a role in the metabolism of antisense drugs. Also underfined are the effects of specific base composition, length, chemical modifications of an oligo, and cellular parameters such as cell type, cell cycle phase and differentiation stage (Mirabelli et al, p. 651).

The post filing date art further confirms the unpredictability of this area. Puri et al (J. Biol. Chem. (2001) 276(31) :28991-28998) teaches "However, despite 40 years of

research, there remain a number of impediments to the successful employment of TFOs as gene targeting reagents. Some of these obstacles reflect the properties of the oligonucleotides. Depending on the nature of the target either purine or pyrmidine TFOs can be used, but there are problems associated with each motif. Under physiological conditions purine TFOs are often subject to self structure formation which is incompatible with triplex formation. (see page 28991, column 2)". Thus, Puri expressly notes that years after Applicant's invention, the invention was still unpredictable. In fact, Puri finds that the nucleotides must be modified in a way not suggested by the application in order to achieve efficacy in what is an ex vivo assay. The complications involved in an in vivo assay would be significantly greater.

Lin et al (J. Biol. Chem. (2000) 275(50) :39117-39124) further supports the unpredictability of this art, noting that "We find that preformed triplexes on DNA that replicated following transfection are less stable than would be predicted by analyses of triplexes in vitro or on total transfected DNA (page 39118, column 1)". The entire gist of the Li paper is that triplex formation ex vivo, in cells, is dramatically different and unpredictably different from triplex formation in vitro. These differences are magnified when compared to in vivo in animal experiments, where issues of delivery, penetration, and other similar issues become relevant.

Quantity of Experimenation

The quantity of experimentation in this area is extremely large since there is significant number of parameters which would have to be studied to apply this technology to in vivo methods, including the stability of the oligonucleotide complex in blood and tissues, the distribution of oligonucleotides in tissues, the optimum mode of effective administration and the pharmocokinetics of administration. For an

oligonucleotide complex, one must also consider (a) the ability of the oligonucleotide to specifically bind the target gene; (b) formation of a stable triple complex between the oligonucleotide and the target gene (note that modification of the oligonucleotide may interfere with its ability to form stable hydrogen bonds, etc.; (c) uptake of the oligonucleotide by the cell; (d) solubility of the oligonucleotide of the cell, and other such constraints. The time table necessary to achieve efficacious administration of effective oligonucleotides, effective temperatures and pH conditions would require a very large quantity of experimentation for in vivo applications. This would require years of inventive effort, with each of the many intervening steps, upon effective reduction to practice, not providing any guarantee of success in the succeeding steps.

Working Examples

The specification has no working examples of in vivo site directed mutagenesis using an oligonucleotide-mutagen complex. While there are in vitro and ex vivo examples, there are no in vivo working examples.

Guidance in the Specification.

The specification provides no evidence that the disclosed effective oligonucleotide-mutagen complexes would be able to modulate nucleic acid interactions or have usefulness in sequence specific triplex formation in vivo, let alone in humans or in a living animal or in plants. The guidance provided by the specification amounts to an invitation for the skilled artisan to try and follow the disclosed instructions to make and use the claimed invention. There is no guidance which suggests how high level recombination could be achieved in animals.

Level of Skill in the Art

The level of skill in the art is deemed to be high.

Conclusion

In the instant case, as discussed above, in a highly unpredictable art where the oligomer-mutagen complexes effects in vivo depend upon numerous known and unknown parameters such as the metabolism specific to the target DNA, potential secondary structure, oligonucleotide length and oligonucleotide chemical composition for triplex DNA, the factor of unpredictability weighs heavily in favor of undue experimentation. Further, the prior art and the specification provides insufficient guidance to overcome the art recognized problems in the use of the oligonucleotide complexes for in vivo treatment as broadly claimed (i.e encompassing a method in any cell under any treatment in any conditions). Thus given the broad claims in an art whose nature is identified as unpredictable, the unpredictability of that art, the large quantity of research required to define these unpredictable variables, the lack of guidance provided in the specification, the absence of a working example and the negative teachings in the prior art balanced only against the high skill level in the art, it is the position of the examiner that it would require undue experimentation for one of skill in the art to perform the method of the claim as broadly written.

Conclusion

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Jeffrey Fredman whose telephone number is 703-308-6568. The examiner can normally be reached on 6:30-4:00.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Gary Benzion can be reached on 703-308-1119. The fax phone numbers for the organization where this application or proceeding is assigned are 703-305-3014 for regular communications and 703-305-3014 for After Final communications.

Any inquiry of a general nature or relating to the status of this application or proceeding should be directed to the receptionist whose telephone number is 703-308-0196.

Jeffrey Fredman
Primary Examiner
Art Unit 1634

July 23, 2003